Microfluidic Implementation of Electrophoretic Fractionation Techniques for the Detection of Biological Particles

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Detection of life on other planets poses unique challenges: since literally nothing is known about theoretical modes of xenobiotic life, techniques must be developed that allow non-specific methods of sample fractionation. In addition, these techniques must be sufficient flexible to permit alteration of fractionation parameters as we learn more about the characteristics of the sample. All *known* biological particles (e.g., bacteria, eukaryotic cells, and proteins) have charged surface groups, which lead to an overall zeta potential for the particles. Of these particles, a large fraction are amphoteric - the zeta potential can be either positive or negative depending on local buffer conditions; under certain conditions the particle will be neutrally-charged. We are working to exploit these phenomena by developing microfluidic devices that can rapidly and continuously fractionate a heterogeneous mixture of particles and then concentrate target particles. Current device designs are targeted towards improving detection of airborne biological and chemical warfare agents through preconditioning a sample stream exiting an air sampler. Both sedimentation and electrophoresis are used to isolate and concentrate particles of interest from interferent particles, such as dust and pollen, to improve the efficiency of downstream analysis.

These techniques are well suited to rapid isolation and detection of biological particles, both known and unknown, particularly when confronted with a real-life heterogeneous sample. Targeted sedimentation modules can continuously fractionate particles based on density or simply remove interferent particles that would clog downstream channels. Electrophoresis and/or isoelectric focusing modules can be used to fractionate and concentrate classes of particles based on zeta potential. These processes could be the basis of a first-pass, non-specific screen for unknown biologics in extraterrestrial samples. Based on the results of these screens, voltages and flowrate can easily be modified to change the range and specificity of the zeta potential selection.

The term "microfluidics" refers to a rapidly growing area of research into what are essentially pipes in which at least one dimension is less than 1mm. In such devices, fluid flow remains almost exclusively in the laminar regime and viscous forces predominate over inertial forces. At these low Reynolds number conditions, convective mass transport between adjacent fluid steams does not occur. Rather, cross-stream mass transport is mediated by diffusion and by movement of particles in applied fields (e.g., electrical, magnetic, gravitational). The majority of research into implementation of electrophoresis in microscale devices has focused on capillary electrophoresis, in which the electric field is parallel to the direction of fluid flow and samples are processed in batch mode. However, our group is currently working on implementing zone electrophoresis (ZE) and isoelectric focusing (IEF) in microfluidic devices. In both ZE and IEF, the electric field is applied perpendicular to the direction of fluid flow. This change in geometry allows continuous sample processing while presenting several challenges in device construction.

Microfluidic devices are particularly amenable to electrophoresis-based applications; the small channel dimensions allow one to generate electric fields on the order of 25 V/cm while keeping the applied voltage low (less than 1.2V). Given a typical electrophoretic mobility of ~1μm/s/V/cm, the resulting terminal velocity of 25μm/s allows a particle to travel across a 500μm channel in only 20 seconds. By using such a small voltage, energy consumption is reduced and gas bubble production at the electrodes is minimized or even eliminated. Other benefits of using microfluidic technologies include reduced power consumption, as well as required reagent and sample size. As fabrication technologies continue to improve, multiple components (e.g., pumps, MEF units, detection units) can be integrated into a single device, which will reduce the overall device size and minimize problems with interconnects.

We have demonstrated concentration of bacteria under flowing conditions in microelectrophoretic (MEF) devices. We have also demonstrated the electrolysis-mediated generation of a pH gradient in similar devices (ampholytes not required). Based in part on those results, we are developing a model of the electrochemical phenomena that occur in MEF devices. This model incorporates diffusion, electroneutrality constraints, multiple electrode reactions, and equilibrium reactions in the channel.